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B-SCIENCES

SEC—B SCI 31

1- Pollen ultrastructure of Chrysobalanaceae

V. C. Patel, J. J. Skvarla & P. H. Raven.....1

2. Ontogeny of the shoot apex of Trapa bispinosa Roxb

D. K. Srivastava.....11

3. Effect of pretreatment of herbicide Stam F-34 (propanil) on germination and early seedling growth of groundnut

M. P. Reddy & A. B. Vora.....25

4. Complex Es of some metal halides with B-arylaminocrotono-phenylhydrazides

Arvind G. Mehta.....33

5. Potential local anesthetis Part XV synthesis of α- secondarya-mino-N- (*w. w.*- diarylalkyl) acetamides.

M. D. Bhatt, J. J. Trivedi.....39

6. u—Homeomorphisms

Piyush Bhatt & Darshan Singh.....43

Vol. 26] * January-August 1983 * [Nos. 1-2

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POLLEN ULTRASTRUCTURE OF CHRYSOBALANACEAE

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Abstract

Pollen of the Chrysobalanaceae genera *Licania, Couepia,* and *Parinari,* was examined by electron microscopy to assess their morphologic and systematic affinities. With scanning electron microscopy (SEM), short, meridional grooves characterize the mesocolpia of *Parinari* pollen while in *Licania* the colpus margin over the endoaperture is distinctly granular. With transmission electron microscopy (TEM), sections in the equatorial regions of *Licania, Parinari,* and *Couepia* all have an undulate or a zig-zag columellae layer which is less typical of angiosperm pollen.

Introduction

Chrysobalanaceae is a tropical family of trees and shrubs with about 17 genera and 450 species. Although traditionally treated as a subfamily of the Rosaceae, there appears to be sufficient anatomical, morphological, and chemical evidence to separate it from the Rosaceae and it is recognized as a separate family by most recent authors (Prance, 1972 ; Takhtajan, 1980 ; Cronquist, 1981 ; Dahlgren, 1981 ; Thorne, 1981). Chrysobalanaceae also shows some attributes similar to the Myrtales : the flowers are epigynous with the sepals, petals and stamens inserted on the rim of a hypanthium tube ; the embryo most commonly contains protein and fat ; and the seeds are endospermless. The latter feature also occurs in the Rosales. Because of this possible mytralean relationship, pollen of a few species of Chrysobalanaceae was examined by SEM and TEM and contrasted with current pollen studies in the Myrtales (Patel, Skvarla, and Raven, in press).

* Dedicated to Bapu : Professor C. M. Patel who inspired two decades of generations in Botany of Gujarat.

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Materials and Methods

Acetolysed pollen (Erdtman, 1952) was air-dried on a specimen stub, sputter coated with gold, and examined with an ISI Super II scanning electron microscope. For TEM, acetolysed pollen was stained in 0.125% aqueous O_3O_4 and 0.5% aqueous uranyl acetate followed by agar embedding, dehydration, and embedding in Epon-Araldite resin mixture (Skvarla, 1966). Thin sections were cut with a diamond knife on a Porter-Blum MT2–B Ultramicrotome, stained in saturated uranyl acetate solution and lead citrate (Venable and Coggeshall, 1965), and examined in a Philips 200 transmission electron microscope. Glycerin jelly slides (Erdtman, 1952) were observed with transmitted light using a Leitz Ortholux microscope.

Table 1 lists the specimens examined and collection data.

Results

The pollen of *Licania michauxii* Prance (Plate I, A–C) and *Parinari klugii* Prance (Plate I, D) is tricolporate, radially symmetrical, spheroidal to suboblate in lateral view, and triangular to triangular-hexagonal in polar view. The surface is smooth punctate. In *Licania,* punctae are scattered over the entire grain. In *Parinari,* fewer punctae are present on the poles and at the margins of the mesocolpia than on the rest of the mesocolpia. Moreover, in *Parinari,* two or three short meridional grooves are present at the middle of the mesocolpia. The colpi are long, with acute ends and a coarse, granular surface. In *Licania,* the granular colpus membrane persists near the open endoaperture (Plate I, B, arrow). The endoapertures in *Licania* are difficult to observe with light microscopy. They are small, lalongate-elliptic. The endoapertures of *Parinari* are large, lalongate, and appear to be elliptic with a very irregular or disrupted margin.

Pollen of *Couepia edulis* (Prance) Prance is also tricolporate, radially symmetrical, spheroidal to suboblate in lateral view, and hexagonal to circular in polar view. The distinct endoapertures are lalongate-rectangular.

Analysis of the transmission electron micrographs of *Licania* (Plate II, A), *Coupia* (Plate II, B), and *Parinari* (Plate II, C,D) indicates that all typical exine layers are present. The endexine is thick below colpi, thin and disrupted in the mesocolpia. The tectum and foot layer are highly irregular, appearing as opposite and alternating hills and valleys. Because of this great irregularity, the columellae layer separating them is notably

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undulate or zig-zag. The columellae are thin in *Licania* and *Parinari* and thick in *Couepia*. In *Couepia*, the undulate columellae layer is not as clearly delineated as in the other two taxa.

The undulate configuration of the columellae layer appears to be fully developed near the equator as is shown in a near median cross section in Plate II, A. Sections cut at different planes do not always show this morphology, in these a less undulate or straight columellae layer is evident. This is exemplified by Plate II, D.

Discussion

The pollen of Chrysobalanaceae has been studied at the light microscope level by a few authors. Erdtman (1952) examined *Hirtella selloana* Hook. fil. and *Licania* guianensis O. Ktze. and concluded (p. 382) "Pollen morphological objections cannot be raised against regarding Chrysobalanaceae as a separate family". Barth and Silva (1963) described the exines of *Hirtella hebeclada* Moric. and *Licania* sp., reporting the latter as having spinules. Melhem and de Paula (1966) studied the pollen of *Couepia* grandifl ra (Mart. et Zucc.) Benth., *Hirtella americana* Aubl., and *Licania humilis* Cham. et Sclecht. They also clarified the above observation of spinules on pollen of *Licania* sp. by stating (p. 213) "The isolated position of this last "*Licania*" was, later on, dismissed by an erratum added by the authors (Brath and Silva) to a reprint, in which it is stated that the grains of figure 8 (in Barth and Silva, 1963) actually belong to *Cordia sellowiana* Cham., a Borraginaceae". Demchenko (1973) made the most comprehensive study of the family, describing the pollen morphology of *Parinarium brasilense* Hook., *Angelesia splendens* Korth., *Couepia bracteosa* Benth., *Parastemon urophyllum* D.C., *Licania arborea* Seem., *Grangeria borbonica* Lam., and *Lecostemon macrophyllum* Spuruce. Three pollen types were established. (1) *Parinarium* type with short-axial pollen with a very thick exine and a ± distinct columellae layer. *Parinarium brasilense* and *Angelesia splendens* belong to this type which is considered to be the oldest by Demchenko. (2) *Couepia* type with long-axial pollen with mexine (Kuprianova, 1956) forming thickenings around the colpi ends ; this type is based on *Couepia bracteosa* and *Parastemon urophyllum*, and is the intermediate type. (3) *Licania* type with long-axial grains that have a thick exine, reduced columellae layer and mexine thickenings around colpi ends

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not as prominent as in *Couepia* type. This is the most advanced type and includes *Licania arborea* and *Grangeria borbonica*. Demchenko reaffirmed Erdtman's contention that the Chrysobalanaceae, on palynological grounds, should be a separate family. According to Demchenko, the pollen of *Lecostemon* differs from the rest of the family, and thus supports exclusion of that genus from the family as suggested by Schulze-Menz (1964). *Lecostemon*, better known as *Rhabdodendron*, is separated as the monogeneric family Rhabdodendraceae by Prance (Cronquist, 1981).

Although Chrysobalanaceae pollen apertures are usually described as tricolporate, Prance (1965, 1972) suggests that the pollen is without distinct pores. This character was used to help distinguish Chrysobalanaceae from *Stylobasium* (Prance, 1965) and Rosaceae (Prance, 1972). The endoapertures in Chrysobalanaceae pollen are of considerable interest, as indicated earlier (see Results), they are not clearly seen in all taxa, and a review of the studies of others indicates that endoaperutre shape is notably variable.

A considerable amount of uncertainty concerns the narrow columellae layer, which we have described as undulate or zig-zag. It seems to be most prominently developed in the equatorial region as previoulsy discussed. In other regions of the grain it seems to be more uniform or even and consists of very short and thin columellae. Erdtman (1952) considered *H. selloana* as having vacuoles (= columellae) and *L. guianensis* to be without them. Barth and Silva (1963) sketched *H. hebedlada* as clearly having columellae. Melhem and de Paula (1966) did not observe columellae in *C. grandiflora*, questioned their presence in *L. humilis*, and saw them in *H. americana* only with the aid of oil immersion optics. Demchenko (1973) noted distinct columellae in *P. brasilense* and *P. urophyllum* ; reduced columellae in *L. arborea* ; almost imperceptible columellae in *C. bracteosa* and *G. borbonica*; and finally, in *A. splendens* columellae were not in evidence.

The undulate or zig-zag columellae layer is somewhat unusual in angiosperm pollen and at present it has been rarely documented. To our knowledge, it has not been described in Rosaceae pollen (Byatt, 1976 ; Kozar and Aaron, 1976 ; Nilsson, Praglowski, and Nilsson, 1977). It does appear remarkably similar to pollen of *Alzatea* (Patel, Skvarla, and Raven in press), and *Saurauia*, particularly *S. barbigera* (Dickison, Nowicke, and Skvarla, in press). *Alzatea* has an uncertain systematic position ,and has been treated as a monogeneric family Alzateaceae (Dahlgren and Thorne, in press), as

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well as member of the Lythraea (Lourteig, 1965) and Crypteronoaceae (Muller, 1975). In SEM, comparisons with *Alsatea* are also favourable although considerably more generalized. *Alzatea* pollen, like Chrysobalanaceae, has a smooth-punctate surface, but the punctae are more concentrated in the elliptic areas at the middle of the mesocolpia (Patel, Skvarla, and Raven, in press). These areas have been suggested as indicating incip ent or rudimentary pseudocolpi (Muller, 1975). Pseudocolpi are not present in Chrysobalanaceae although the meridional grooves in *Parinari klugii* are not understood at this time. *Saurauia* belongs to the Actinidiaceae and has recently been examined in a comparative pollen study of Actinidiaceae and Dilleniaceae (Dickison, Nowicke, and Skvarla, in press). In SEM, however, *Saurauia* differs from Chrysobalanaceae in being psilate and in having ektexine bridges covering the endoapertures.

Exines having an undulate columellae layer somewhat similar to the Chrysobalanaceae are also present in the Corynocarpaceae (Nowicke and Skvarla, in press). This pollen, however, is dicolporate and syncolpate, and appears related to certain taxa in the Saxifragaceae.

According to Dahlgren (Dahlgren and Thorne, in press), " In the light of the several conspicuous differences between Chyrsobalanaceae and and the Rosales (*s. str.*) it may be premature to consider the family a connecting link between Rosales and Myrtales. The position of Chrysobalanaceae needs careful reconsideration .....the number of similarities with Myrtales (whether indicating affinity or being due to convergent evolution) are indeed noteworthy ". Apart from the similarity to *Alzatea,* a more comprehensive investigation of Chrysobalanaceae pollen is in order before possible Myrtales connection can be conclusively evaluated. Further, pollen studies of those Rosaceae considered near to Chrysobalanaceae would also be useful.

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Acknowledgements

Supported in part by grants from National Science Foundation to J.J.S. and P.H.R.

TABLE 1 Specimens examined.

Texa	*Locality*	*Collector*	*Hebarium*
Licania michauxii Prance	Florida	Foster, Smith & Smith s.n.	OKL
Couepia edulis (Prance) Prance	Brazil	Ramos s.n.	OKL
Parinari klugii Prance	Peru	Croat 20443	OKL

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Plate 1. SEM of Chrysobalanaceae.

Fig. A-C. *Licania michauxii.* A Lateral view. ×2,000. B. Surface details near the endoaperture. The surface is punctate. The colpus membrane has granular to slightly elongated elements on it. They persist near the open endoaperture (arrow). ×7,000. C. Polar view. ×2,800. Fig. D. *Parinari klugii.* Polar view. Note there are few punctae at the pole and near colpi. Arrows indicate meridional grooves. ×2,000. All lines equal 1 um.

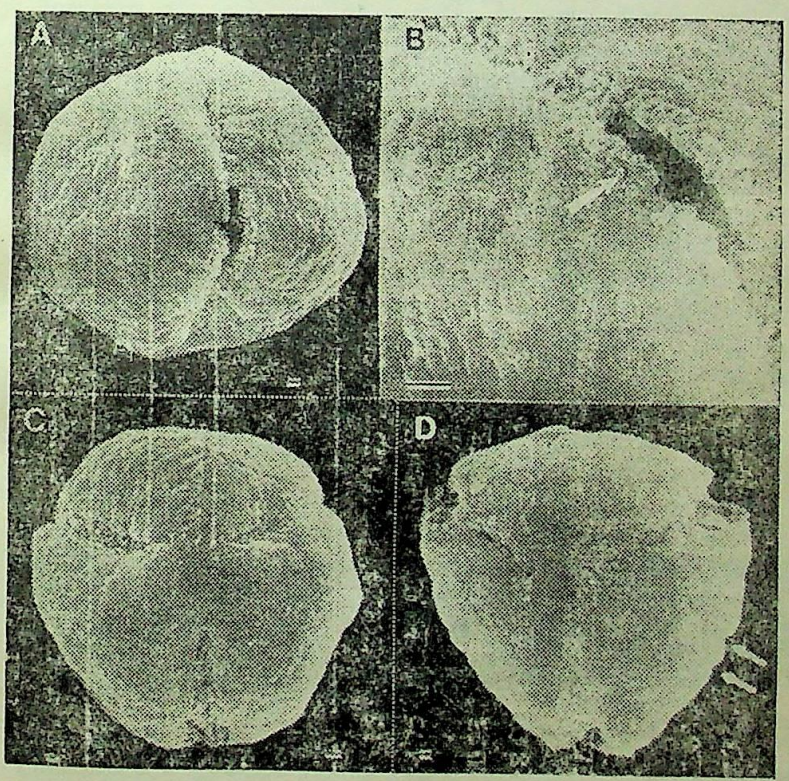

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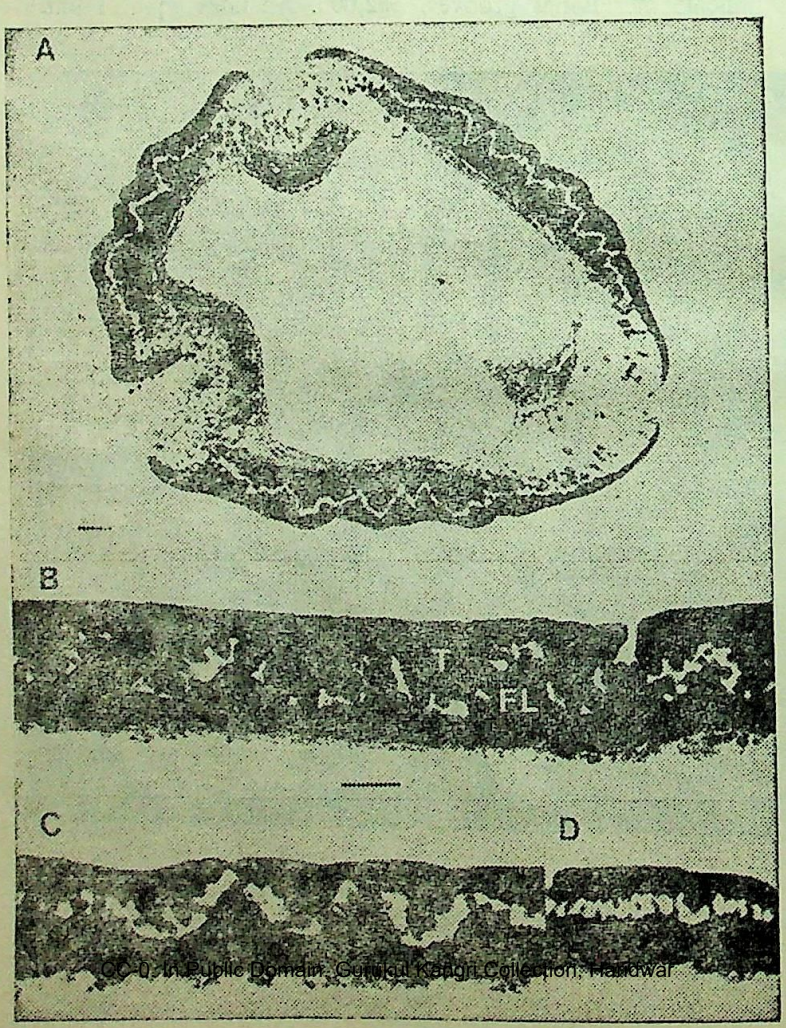
Plate 2. TEM of Chrysobalanaceae.

Fig. A. *Licania michauxii.* TEM of a near median section passing through three colpi. The endexine is thick below colpi, thin and fragmented elsewhere. The tectum and foot layer are thick. Their lower and upper margins respectively, correspond to the undulations of the narrow columellae layer. The columellae are thin and short. ×5,500. Fig. B. *Couepia edulis.* Section of a mesocolpium near the equator. The undulate columellae layer between the tectum (T) and the foot layer (FL) is not as distinct as in Fig. A and C. Here, the columellae are thick and the intercolumellar spaces are small. The endexine is thin and disrupted and is not easily distinguishable due to heavy staining. ×10,000. Fig. C,D. *Parinari klugii.* C. Section of a mesocolpium near the equator. The undulate columellae layer is distinct. ×13,000. D. Section of an area not near the equator. In contrast to Fig. C, the columellae layer is more or less straight, with thin, erect columellae. The tectum is thinner than the foot layer ; the endexine is disrupted. ×7,000. All lines equal 1 um.



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ONTOGENY OF THE SHOOT APEX OF *TRAPA BISPINOSA* ROXB.

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Abstract

Quantitative changes in RNA, total proteins and —SH protein have been measured in meristematic cells of the shoot apices viz. embryonal, seedling, vegetative and reproductive. The growing apex imprints a biseriate tunica, corpus mother cell zone, peripheral zone and pith rib-meristem. Different zones depict different stain intensities. The cells of the pith rib meristem are vacuolate, faintly stained and show highest cell area. Insoluble polysaccharides are studied qualitatively. Except for the tunica other zones of the shoot apex show uniform cell wall thickening with PAS reaction. Starch grains pile up at the base of the leaf primordium. The cell area of different zones of the vegetative shoot apex at the instance of floral induction is less as compared to the vegetative stage. A significant rise in the diameter of the apical dome and cell number in different zones are simultaneous with the floral induction. Different zonations of the vegetative shoot apex show high extinction values for RNA and total proteins but less for —SH protein, while a reverse trend at floral induction. A single layered tunica occurs in floral, stamen and carpel primordia. These show dense stain for all the metabolites. The axillary bud turns vegetative and forms branches in the older leaves. The embryonic shoot apex and developing primordia show uniseriate where as the vegetative shoot apex shows biseriate tunica at the vegetative phase and at floral induction. The embryonal shoot apex arises as in monocotyledons and has a single layered tunica with cells smaller than the corpus.

DKS acknowledges the financial assistance from UGC.

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Introduction

Histochemical detection of different metabolites in angiosperm shoot apical meristem revolves round only a few studies (Gifford and Tepper, 1962; Nougarede, 1967; West and Gunckel, 1968 a, b; Bernier, 1971; Gifford and Corson, 1971; Evans and Berg, 1972; Shah et al. 1974; Shah and Raju, 1975 ; Shah and Jain, 1980). The shoot apex is a multicomponent system in which its different sectors continually interact. It normally produces leaves, axillary buds and reproductive organs, so diverse cell types develop from the meristem to serve various physiological functions. On the basis of staining reaction Foster (1958) in gymnosperms and Boke (1941) and Gifford (1950) in angiosperms distinguished cyto-histological zonations encompassing both tunica and corpus. Many workers have shown that tunica and corpus are two very distinct zones supporting cytohistological zonations. In *Trapa* the embryonic shoot apex continues to exist as a vegetative one. The reproductive or the floral apex resides in the axil of a leaf. The present investigation studies the form-histochemistry of the shoot apices and behaviour of foliar and floral apices in *Trapa*, an economic plant. Fruits of *Trapa* are valuable food in India.

Material and Methods

The plants of *Trapa* were collected from the Chandola lake (10 k.ms. away from Ahmedabad) and grown in the ponds of Gujarat University Botanical garden. The shoot apices were fixed in Carnoy's fluid for 1-1½ hrs. and sectioned at 10 microns. The quantitative study viz. RNA, total proteins and —SH protein were carried out on median sections of three different shoot apices embryonal, foliar and floral. The insoluble polysaccharides were studied qualitatively. Ribonucleic acid (RNA) was demonstrated by pyronin (G) at pH 3.5 (Tepper and Gifford, 1962), proteins by mercuric bromophenol blue (HgBPB) (Mazia et al., 1953) and-SH protein by DDD and fast blue and insoluble polysaccharides with PAS (Jensen, 1962). The relative quantitative determinations of cell constituents and stain intensities were measured with the help of cytophotometer using narrow band filters. We have taken the reading of 10 cells and 10 readings from each cell from different zonations of the shoot apex as well as developing primordia of leaf, flower, stamen and carpel. Adequate controls were run all the time.

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Observations

Morphology : The inception of the shoot apex occurs at first indentation on the germinal face of the elongated embryo. The shoot apex is lateral in origin. The crevice or groove delimits the shoot apex originating on the lateral face of the embryo precisely as in monocotyledons (Fig. 1A). The embryonic shoot apex of *Trapa* consists of a single layered tunica and huge island of corpus cells (Fig. 1B). No zonations exist within the corpus. The nascent shoot apex has single layered tunica with cells smaller than the corpus.

The vegetative shoot apex is a dome covered by young leaves. Its meristematic cells are thin walled and unvacuolated except pith rib meristem. The growing apex gives rise to the leaf primordia alternately on both sides of the apex by forming a subterminal ridge. It shows cytohistological zonations too (Fig. 1C) i. e. a biseriate tunica (T_1 and T_2), corpus mother cell zone, peripheral zone, and pith rib meristem. The cells of the T_2 are larger than those of the T_1. The cell division in T_1 is strictly anticlinal while T_2 shows frequent periclinal divisions at the site of leaf intiation. The cells of the corpus are large and divide in various planes. The corpus mother cell zone merges laterally with the peripheral zone. The cells of the peripheral zone divide both anticlinally and periclinally and they are slightly smaller than the corpus mother cell zone. Pith rib meristem has vacuolated, big, lightly stained cells arranged in 9–11 vertical files. It is quite distinct in both vegetative and reproductive phase of development.

The first sign of leaf initiation is by periclinal divisions in 3–4 cells of the second tunica. The leaf buttress is formed due to periclinal division in T_2 and corpus, and anticlinal in T_1 on the flank. The flowers in *Trapa* are axillary and acropetal. In older leaves the axillary bud turns vegetative and forms branches. During the reproductive phase the floral primordia develop in the axil of leaves. Soon after a little elongation (Fig. 1D) two centres of growth arise to develop into two sepals. The stamen primordia resemble the floral one where as the carpel primordia resemble the leaf primordia. A single layered tunica is observed in the primordia of flower stamen and carpel. The cells are small and meristematic and show dense stain.

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Cell Area (μ^2) : The embryonic shoot apex consists of single layered tunica and corpus. The cells of tunica (42.84 μ^2) are smaller than those of corpus (59.78 μ^2). The cell area of different zonations of shoot apex varies at vegetative and reproductive phases. The average cell area of tunica, corpus, peripheral zone and pith rib meristem is 52.76μ^2, 83.25μ^2, 60.58μ^2 and 122.36μ^2 respectively during the vegetative phase. During the reproductive phase the mitotic activity increases in the shoot apex. The cell area of tunica, corpus, peripheral zone and pith rib meristem are 37.52 μ^2, 57 12μ^2, 51.38μ^2 and 78.90 μ^2 respectively (Fig. 2A). The cell area of leaf primordia is more or less same at both the phases of development. The cell area of floral, stamen and carpel primordia are 52.17μ^2, 45.20μ^2, and 41.40 μ^2 respectively (Fig. 2A).

Vegetative Shoot Apex

RNA : Pyronin (G) for RNA stains both nucleolar as well as cytoplasmic RNA. The tunica and the peripheral zone show high extinction values for RNA as compared to the corpus mother cell zone. The pith rib meristem shows faint staining (0·0346). The e-value of leaf primordia is quite high (0·0588) (Fig. 2B). The content per cell of RNA is more in the corpus mother cell zone and pithrib meristem but it is extremely less in the tunica and peripheral zone. RNA content per cell of leaf primordium is 2·39 (AU) (Fig. 3A).

Total Proteins : The use of mercuric bromophenol blue for total proteins reveals that e-value and content per cell of total proteins vary in different zonations of the shoot apex. The e-value of tunica is high (0.1370) as compared to the pith rib meristem (0.0680) The e-values of corpus mother cell zone and peripheral zone is more or less same (i e. 0.1211 and 0.1230 respectively) (Fig. 2C). The developing leaf primordium shows maximum e-value of 10·1317. The content per cell of total proteins is high in the corpus mother cell zone (10.08 AU) as compared to the tunica, peripheral zone and pith rib meristem which contain 6 89 (AU), 7.45 (AU) and 8.32 (AU) respectively (Fig. 3B). The content per cell of total proteins in leaf primordium is 5.37 (AU).

–SH Protein : The e-value of –SH protein is high in the tunica, corpus mother cell zone and peripheral zone as compared to the pith rib meristem (Fig. 2D). The e-value in the leaf primordia is 0.0663.

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The content per cell of –SH protein is more in the corpus mother cell zone and pith rib meristem, where as it is less in the tunica and peripheral zone 3.85 (AU) and 4.07 (AU) respectively (Fig. 3C). The content per cell of leaf primordium is 2.70 (AU).

Insoluble Polysaccharides : Among different zonations of the shoot apex the cells of the tunica layer are dark stained with PAS reaction (+++). The cells of the corpus mother cell zone and peripheral zone stain uniformly (++), where as the cells of the pith rib meristem(+) stain faintly. Except for the tunica layers other zones of the shoot apex show uniform cell wall thickening with PAS (Fig 1C). Starch grains are abundant at the base of the leaf primordium. Vascular strands of the shoot apex and leaf primordium show deep stain with PAS reaction.

Vegetative Shoot Apex During Floral Evocation (Reproductive Phase) :

RNA : Pyronin (G) for RNA demonstrates the high e-values in tunica and peripheral regions, whereas the e-values of corpus mother cell zone and pith rib meristem are low (0.0383 and 0.0299 / respectively) (Fig. 2B). Different zonations of the vegetative shoot apex at floral induction show low extinction values as compared to the vegetative one. The e-value of leaf primordia is 0.0441. The developing floral, stamen and carpel primordia show more or less high values for RNA (Fig. 2B). The content per cell of RNA in the tunica is less (1.77 AU) whereas the corpus mother cell zone, peripheral zone and pith rib meristem contain 2.19 (AU), 2.10 (AU) and 2.35 (AU) respectively (Fig. 3A), The content per cell of RNA in the leaf primordia 2.15 (AU). The floral, stamen and carpel primordia show 2.54 (AU), 2.28 (AU), and 2.16 (AU) content per cell respectively. Both nucleolar as well as cytoplasmic RNA abound in the shoot apex and the developing primordia.

Total Proteins : The tunica, corpus mother cell and peripheral zone show high extinction values for total proteins (0.1116, 0.1051 and 0.1154 respectively) whereas the cells of the pith rib meristem stain faintly (0.0740). The e-value of leaf primordia is 0.1286. The floral primordia show little higher extinction value (0.1386) than the stamen and carpel primordia (0.1005 and 0.1053 respectively, Fig 2C). The content per cell of total proteins is more in corpus mother cell, peripheral zone and pith rib meristem, where as it is less in the tunica. The content per cell in

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the leaf primordia is 6.27 AU. The floral primordia show high content per cell than the stamen and carpel primordia (Fig. 3B).

-SH Proteins : The absorbance values of -SH proteins in different zonations of vegetative apex is higher in the reproductive phase than the vegetative one, thereby indicating an increase in the activity. The e-value of -SH protein is high in the corpus mother cell zone (i.e. 0.0780) as compared to the tunica, corpus and pith rib meristem (0.0748, 0.0697 and 0.0490 respectively). The e-value of leaf primordia is 0.0621. The floral, stamen and carpel primordia show high extinction values i.e. 0.0879, 0.0829 and 0.0840 respectively (Fig. 3D). Content per cell of -SH protein is maximum in the corpus mother cell zone (4.45 AU) than in the tunica, peripheral zone and pith rib meristem (2.80 AU), 3.58 (AU), 3.86 (AU) respectively. The content per cell of -SH protein in the developing leaf, floral, stamen and carpel primordia are 3.30 (AU) 4.58 (AU), 3.73 (AU) and 3.47 (AU) respectively (Fig. 3C).

Insoluble Polysaccharides : The periodic acid Schiff's staining reveals the presence of insoluble polysaccharides in different zones of the shoot apex and developing primordia. Except for the tunica layers other zones of the shoot apex show uniform cell wall thickening with PAS (Fig. 1D). The developing primordia of leaf, flower, stamen and carpel show uniform cell wall staining, PAS positive cytoplasm is also observed in different zonations of shoot apex and developing primordia.

Discussion

The shoot apex in *Trapa bispinosa* exhibits a cytohistological zonation as described earlier for many angiosperms (Gifford, 1954; Tapfer, 1960). Based on differential staining and planes of cell division the vegetative shoot apex is delimited into four zones 1. tunica, 2. corpus mothes cell zone, 3. peripheral zone and 4. pith rib meristem. zone. The tunica is is biseriate (T_1 and T_2) and divisions are predominantly anticlinal. Periclinal divisions occur in the T_2 at the time of development of leaf primordia (Evans and Berg, 1972; Trivedi et al. 1980) followed by divisions in underlying corpus cells which increase the girth. In contrast in *Glycine max* the leaf originates exclusively from corpus inspite of the presence of a biseriate tunica (Sun, 1957).

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The reproductive or floral apex is initiated in the axil of a leaf. Single layered tunica is observed in the floral primordium. A rise in the diameter of the apical dome and cell number are observed in the vegetative shoot apex simultaneous with the floral induction. A similar trend was observed by Orr (1981).

Different zones of the shoot apex depict different stain intensities. An ultrastructural stereological study showed that each zone has its own distinct structure (Mauseth, 1981). Extinction values for protein bound sulfhydryl groups remain high in different zones of the vegetative shoot apex during the reproductive phase. Different zones of the vegetative shoot apex show high e-value and content per cell of RNA and total proteins and a decline in the main vegetative apex during floral induction. With the decrease in the e-values and content per cell of these metabolites in the vegetative apex during reproductive phase, there is a simultaneous increase in the floral apex.

West and Gunckel (1968) localized starch in the central zone and pith rib meristem in *Brachychiton* but no starch in the flank cells. In *Trapa* starch grains pile up at the base of the leaf primordium. Few starch grains are present in the pith rib meristem. Except the tunica other zones of the shoot apex show uniform cell wall thickening with PAS reaction.

Embryonic shoot apex shows single layered tunica, whereas the vegetative shoot apex during the vegetative and reproductive phases show biseriate tunica. Floral, stamen and carpel primordia show single layered tunica. Fluctuations in the tunica layers are also observed during the vegetative to reproductive shoot apex in bajra (Daniel and Shah, 1971) and rice (Jain, 1981).

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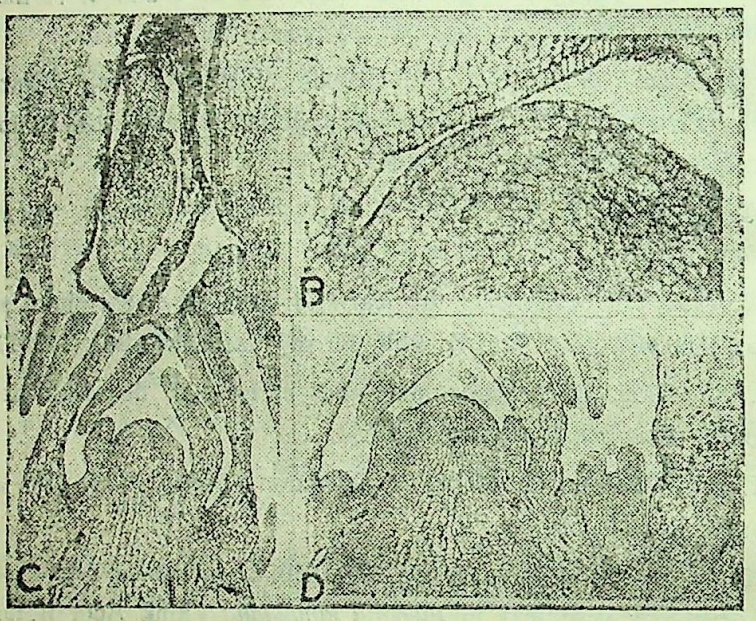
Legend to the figures

Fig. 1 A–D :



Trapa bispinosa, Ontogeny of the shoot apex.

(*A*) Median L. S. of indented embryo stained for insoluble polysaccharides. Note the origin of the shoot apex on the lateral face (germinal face) of the embryo. x 100.

(*B*) Median L. S. of embryonal shoot apex shows single layered tunica and huge mass of irregularly lying corpus cells stained with PAS reaction. x 400.

(*C*) Median L. S. of vegetative shoot apex stained for insoluble polysaccharides shows cytohistological zonation. x 100.

(*D*) Median L. S. of vegetative shoot apex during floral evocation. Note the increase in the diameter of apical dome and cell number in different zones. The reproductive or floral apex arises in the axil of a leaf. Note intense staining of tunica layers with PAS reaction. × 100.

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Fig 2 A. D. :

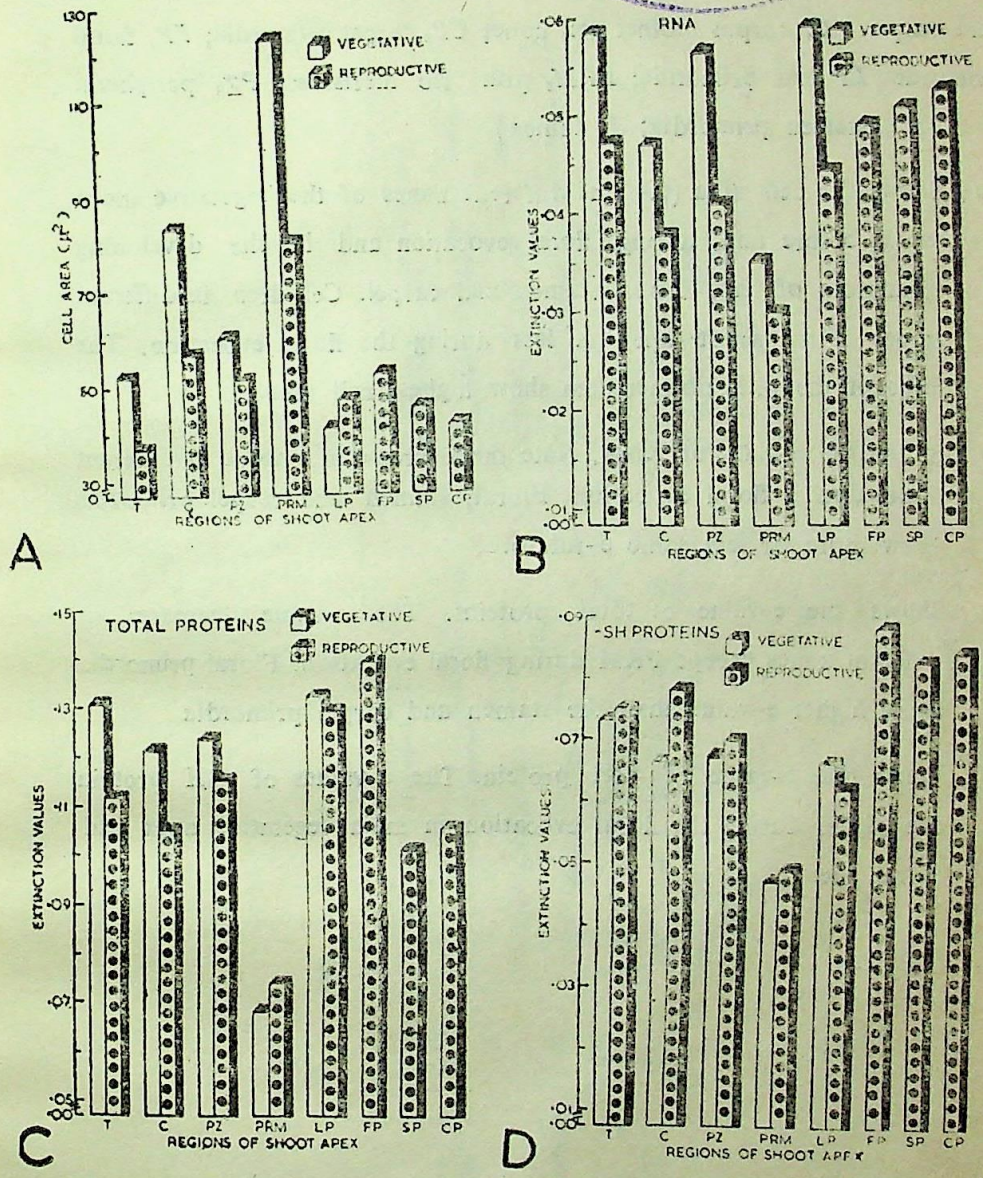

FIG. 2 A–D

Cytological and cytochemical changes in the shoot apex of *Trapa* during the vegetative and floral evocation. Blank histograms show different zones of the vegetative shoot apex where as the dotted shows

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the same during floral evocation and primordia of leaf, flower, stamen and carpel. (*C*, corpus mother cell zone; *CP*, carpel primordia; *FP*, floral primordia; *LP* leaf primordia; *PRM*, pith rib meristem; *PZ*, peripheral zone; *SP*, stamen primordia; *T*, tunica).

(*a*) Shows the cell area (μ^2) in different zones of the vegetative shoot apex before and during floral evocation and in the developing primordia of leaf, flower, stamen and carpel. Cell area in different zones of the shoot apex is less during the floral evocation. The cells of the pith rib meristem show highest cell area.

(*b*) Shows the e-value of RNA. Note the decrease in e-value in different zones during floral evocation. Floral, stamen and carpel primordia show more or less same e-values.

(*c*) Shows the e-value of total proteins. The e-value decreases in different zones except PRM during floral evocation. Floral primordia show higher e-value than the stamen and carpel primordia.

(*d*) Shows the e-value of -SH protein. The e-values of -SH protein are higher during the floral evocation in main vegetative apex and floral apex.

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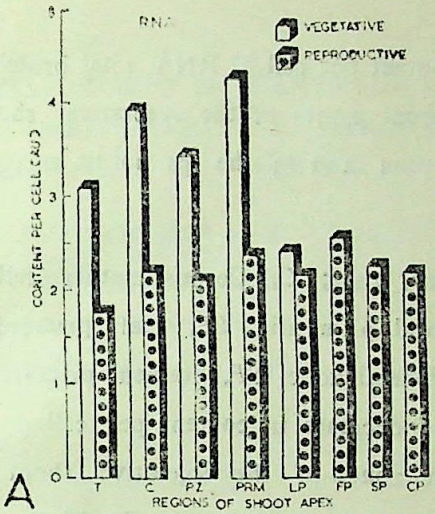
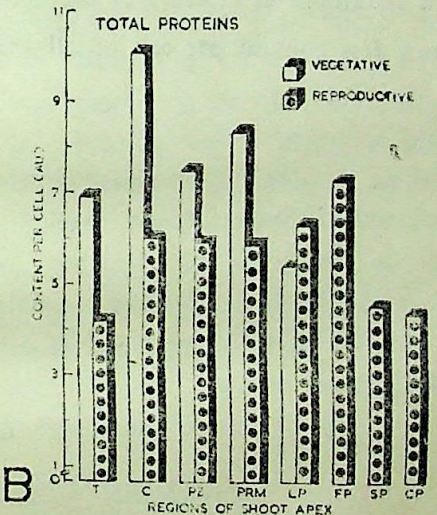
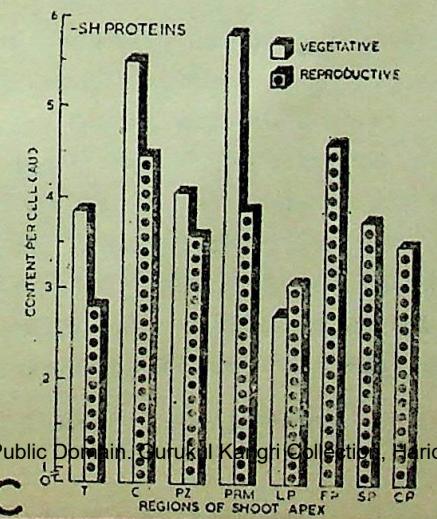


A



B



C

Fig. 3 A–C :

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Shows the content per cell of RNA, total proteins and -SH proteins in AU in the different zones of the vegetative shoot apex before and during floral evocation and in the primordia of leaf, flower, stamen and carpel.

(*AU*, arbitrary, units; *C*, Corpus mother cell zone; *CP*, Carpel primordia; *FP*, floral primordia; *LP*, leaf primordia; *PRM*, Pith rib meristem; *PZ*, peripheral zone; *SP*, Stamen primordia; *T*, Tunica)

Note with the decrease in content per cell of all the metabolites (A-C) in different zones of the vegetative shoot apex during floral evocation; there is a simultaneous increase in the floral apex. Stamen and carpel primordia show less content per cell of all the metabolites.

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EFFECT OF PRETREATMENT OF HERBICIDE STAM F-34 (PROPANIL) ON GERMINATION AND EARLY SEEDLING GROWTH OF GROUNDNUT

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Summary

Seeds of groundnut (*Arachis hypogeae* L. G-10) were surface sterilized with ($HgCl_2$) mercuric chloride and pretreated with different concentrations (10, 50, 100, 200 ppm.) of Stam F-34 (Propanil) for 16 hours Later they were germinated in petridishes lined with Whatman No. 1 filter paper. Germination and early seedling growth was observed. All the concentrations of herbicide showed an inhibitory effect over control. Ove all growth of the seedlings (epicotyl, hypocotyl and radicle) was retarded though the germination was not affected. The effect was more in 100 and 200 ppm Delayed germination, however was observed with increasing co centrations.

Introduction

Several Herbicides are known to bring reduction in growth and germination. They are potential inhibitors of photosynthesis (Astan and Crafts, 1973) 2-4-5-T and Amitral were inhibitory to seedling growth of *Urginia indica* (Khare, and Dubey, 1979). Inhibition of leaf growth and increase in thickness of the leaf was observed in groundnut and sunflower with H-33 (2-6 Dichlorobenozitrile) (Koopman and Damas, 1960) swelling of stem, promotion of axillary buds was observed with Piclorm and TCA with high concentrations and significant promotory effect in low concentrations was also observed (Fisher et al. 1968). Barban and Pro-

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pachlor retarded the growth and germination of grass seeds (Mannan et al 1965).

With these points in view the effect of Stam F-34 (Propanil) on the germination and early seedling growth of groundnut (*Arachis hypogeae* L) was studied.

Materials and Methods

Groundnut (*Arachis hypogeaea* L. G-10) seeds of uniform size were surface sterilized with 0.1% ($HgCl_2$) mercuric chloride for 30 mints, thoroughly washed and were pretreated with different concentrations (10, 50, 100, 200 ppm) of Stam F-34 (Propanil) for 16 hours. Later they were germinated in petridishes (dia. 9 cm.) having Whatman No 1 filter paper. Each petridish contained 5 seeds. Experiments were carried out at room temperature Water was replaced in petridishes at regular intervals (12 hours); growth data was taken at 2 days intervals upto 10th day (4-10) separately (Radicle, Hypocotyl and Epicotyl). Four petridishes were sampled every time from each treatment when the seedlings were of appropriate age. Embryo axises were separated into radicle hypocotyl, epicotyl and their length was measured Fresh weight, dry weight of cotyledons and embryo axis were also taken. From this data water content of individual parts was calculated.

The experiment was repeated four times and the constant readings were observed,

Results and Discussion

Herbicides are wellknown inhibitors of germination and growth. However, germination was not affected in this experiment. Slight delay in germination was observed.

The first step for the germination of seed is the hydration of cotyledons and embryoaxis. Results in table IV prove that hydration in cotyledons and embryo axis decreased with increasing concentration of herbicide and may be because of high osmatica which causes delay in mobalization of reserve food materials from cotyledons to embryo axis.

Results presented in table I show that the length of the hypocotyl, epicotyl and radicle decreased with increasing concentrations, Epicotyl was more affected. The overall reduction in growth including increment

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in fresh weight seems to be due to the reduced translocation of reserve food materials from cotyledons to embryo axis as the dry weight of the cotyledon remain higher under high concentration of herbicide treatment The effect on embryo axis is opposite to that of cotyledons (Tables II, III).

Inhibitory effect of root elongation and swelling of hypocotyl was observed. The results shpport the data of Ashtan et al., (1969). The swelling in hypocotyl may be due to stimulation in cambial activity in the hypocotyl as reported in bean plant treated with Dichobenil (Koopman and Dammas, 1960), reduction in leaf size and increased in leaf thickness were supporting the result of Koopman and Dammas, 1960).

Thus the symptoms of herbicides are similar to those of stress.

Aknowledgement

The receipt of CSIR fellowship to M.P.R. is greatfully acknowledged.

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TABLE I

Effect of pretreatment with different concentrations of Stam F-34 (Propanil) on hypocotyl, epicotyl and radicle length of groundnut (cm.)

Preatments (ppm)	*Days after germination*								*Epicotyl on 10th day*
	Hypocotyl				*Radicle*				
	4	6	8	10	4	6	8	10	
Control	1.6 ±0.05	2.6 ±0.09	3.0 ±0.11	3.2 ±0.13	2.1 ±0.07	3.1 ±0.09	3.4 ±0.13	3 6 ±0.15	6 0 ±0.18
10 ppm	1.6 ±0.06	2.3 ±0.11	2.9 ±0.08	3.1 ±0.04	2.1 ±0.07	3.1 ±0.08	3.3 ±0.14	3.6 ±0.13	5.5 ±0.14
50 ppm	1.6 ±0.05	2.1 ±0.08	2.3 ±0.09	2.5 ±0.11	1.6 ±0.06	2.6 ±0.08	3.0 ±0.11	3.3 ±0.14	5.1 ±0.09
100 ppm	1.4 ±0.07	1.7 ±0.09	1.8 ±0.07	1.9 ±0.09	1.2 ±0.08	2.1 ±0.11	2.5 ±0.09	2.9 ±0.15	3.0 ±0.05
200 ppm	0.7 ±0.03	1.2 ±0.07	1.3 ±0.06	1.5 ±0.09	1.1 ±0.05	1.4 ±0.07	1.9 ±0.11	2.3 ±0.09	2.2 ±0.09

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TABLE II

Effect of pretreatment with different concentrations of stam F–34 (Propanil) on fresh weight of cotyledons–embryo axis of groundnut (values are S.D. ± mg/seed)

Treatment (ppm)	Days after germination									
	Cotyledons					Embryo axis				
	2	4	6	8	10	2	4	6	8	10
Control	645.00 ±9.03	708.34 ±11.13	785.35 ±12.65	820.11 ±17 62	255.19 ±20.32	480 00 ±8.42	995.31 ±9.63	1385.0 ±16.85	1651.35 ±21.45	1775.15 ±28.45
10 ppm	645.73 ±10.75	700.00 ±12.05	769 45 ±14.73	808.21 ±16.32	815.70 ±19.75	481.35 ±3.54	976.63 ±10.32	1370.35 ±16.75	1637.00 ±23.12	1758 00 ±27.85
50 ppm	642.03 ±10.64	700.45 ±13.21	765 00 ±15.32	805 31 ±15.32	820.05 ±20.65	465.73 ±6.12	974.37 ±9.62	1355.00 ±19.95	1614.09 ±25.45	1756.43 ±26.05
100 ppm	635.45 ±9.65	689 00 ±9 65	730.37 ±16.35	785 09 ±19.35	850.00 ±20.32	435.73 ±8.62	905.43 ±9 65	1252.00 ±19.35	1501.39 ±23.05	1688.51 ±29.47
200 ppm	638.00 ±9.64	663.45 ±11.63	705.45 ±16.45	765.00 ±19.75	865.45 ±21.32	405.03 ±7.64	865 51 ±8 69	1203.09 ±20.35	1381.41 ±28.43	1521.05 ±29 08

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TABLE III

Effect of pretreatment with different concentrations of stam F-34 (propanil) on dry weight of cotyledons—embryo axis of groundnut (values are S.D± mg/seed)

Treatment (ppm)	*Days after germination*									
	Cotyledons					*Empryo axis*				
	2	4	6	8	10	2	4	6	8	10
Control	410.45 ±5.75	360 34 ±5.05	319.64 ±4.11	285.64 ±3.00	235.73 ±4.07	30.37 ±1.54	95.45 ±2.21	125.43 ±3.05	201.74 ±3.43	265.39 ±4.64
10 ppm	405.70 ±6.37	365.03 ±4 95	331 05 ±4.07	305.35 ±3 17	285.05 ±5.05	31.75 ±1.74	83.34 ±2.21	117.74 ±2.95	197 35 ±3.05	255.43 ±4.09
50 ppm	406.00 ±5.95	370.37 ±4.75	340.55 ±4.09	315.35 ±4 05	290.00 ±4.00	29.34 ±1.95	85.73 ±2.86	115.11 ±2.93	185.00 ±2.09	251 53 ±4.15
100 ppm	403 79 ±5.35	390.57 ±4.07	350.70 ±4.95	340 95 ±3.75	303.45 ±3.93	20 00 ±1.47	65.13 ±1.95	102.17 ±2.25	151.00 ±3.47	203.45 ±4.05
200 ppm	405.80 ±6.03	393.39 ±4.79	376.40 ±3.09	360.00 ±3.95	335.30 ±4.09	15 43 ±0.95	55.00 ±1.95	95.09 ±2.03	131.73 ±2.95	176 63 ±3.74

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TABLE IV

Effect of pretreatment with different concentrations of Stam F-34 (Propanil) on water content of cotyledons-embryo axis of groundnut (per cent of dry weight bases) (values are S D. ± mg/seed)

Treatment	Days after germination									
	Cotyledons					embryo axis				
(ppm)	2	4	6	8	10	2	4	6	8	10
Control	235.15 ±4.75	348.34 ±8.09	465.75 ±8.05	530.17 ±8 95	520.49 ±8.75	450.11 ±8 75	900.13 ±12.75	1260 45 ±16 32	1450.79 ±19.75	1510.12 ±24.32
10 ppm	240.23 ±8.09	345.00 ±6.75	438.43 ±11.73	503 34 ±10.34	530.37 ±10.34	450.43 ±10.35	893.00 ±13.75	1253.93 ±17.32	1435.19 ±30.73	1503 00 ±26.75
50 ppm	236.57 ±5·32	330.35 ±5 95	425.67 ±6.75	484.54 ±10.75	540.87 ±8.75	439.95 ±9.75	889.71 ±14.75	1240.15 ±20.32	1429.23 ±29.75	1505.73 ±25.32
100 ppm	232.68 ±8.32	300.70 ±6.95	370.95 ±8.47	445.43 ±11.45	547.64 ±9.45	415.36 ±11.34	840.91 ±3 35	1150.34 ±18.32	1350 43 ±21.73	1485 09 ±23.75
200 ppm	233.34 ±6.84	265.07 ±7.65	355.00 ±9.75	405.10 ±8.45	530.30 ±9 73	390.87 ±9 75	810 91 ±15.34	1110.19 ±19.37	1250 59 ±20.35	1345 73 ±19.75

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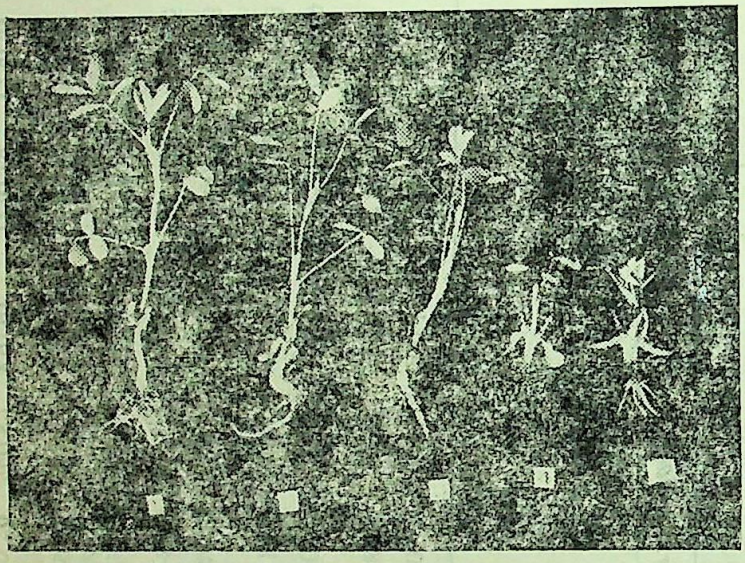

1. Control
2-5 Different treatment of herbicide
2. 10 ppm
3. 50 ppm
4. 100 ppm
5. 200 ppm

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COMPLEXES OF SOME METAL HALIDES WITH β-ARYLAMINOCROTONO-PHENYLHYDRAZIDES

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Abstract

The interaction of β-arylaminocrotonophenylhydrazide (APH) with Cd (II) chloride, Cd (II) iodide, Hg (II) chloride and Zn (II) chloride yields 1 : 1 addition products, which have been characterised by molecular weight and analytical data.

The addition products of β-arylaminocrotonates with Cu(II) and Zn(II) and Cd (II) chlorides have been known.[1] The spectro-photometric study of copper (II) ethyl-β-2-methoxyanilino-crotonate complex indicated 1 : 1 composition of The complex.[2] Desai and Desai[3] prepared complexes of Zn (II) chloride with (APH). Desai, Mehta and Desai[4] prepared complexes of Cd (II) chloride, Cd (II) iodide and Hg (II) chloride with (APH). The present communication deals with the study of complexes of Cd (II), Hg (II), Zn (II) chlorides and Cd (II) iodide with 2-methoxy, 3-methoxy and 4-methoxy anilinocrotonophenylhydrazides.

Experimental

Ethyl β-arylaminocrotonate were prepared by the known method[2].

The equimolar (0.03 M) mixture of crotonate and phenylhydrazine was refluxed on a water bath for half an hour and the reaction mas was leached with 5% hydrochloric acid, saturated sodium chloride and

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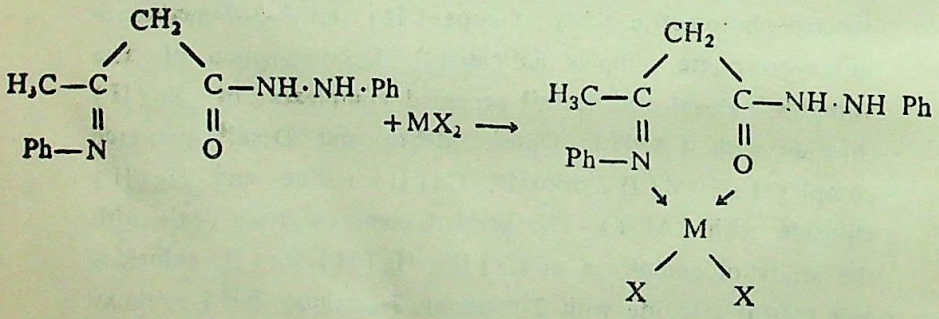
water and then extracted with anhydrous ether; evaporation of ether gave the pure product.

APH (0.03 M) in alcohol (8.0 ml) was treated with ethanolic (15.0 ml) metal halide (0.03 M) solution and stirred well at R.T. for half an hour when fine crystals of the product separated. It was washed with ethanol and ether. The metal and halogen were estimated by the usual methods.

These products are crystalline solids and are insoluble in ether. The complexes of Cd(II) and Zn(II) are soluble in alcohol, while Hg(II) complexes are sparingly soluble in alcohol. They are soluble in hydrochloric acid with decomposition. The analytical data and molecular weight indicate 1 : 1 composition of the complexes. (Table-I).

I R Spectra : In the present study, it has been observed that even if excess of ligand is added to the metal halide, only one mole of the ligand was bound to the metal into the resulting complexes $HgLX_2$, $CdLX_2$ and $ZnLX_2$ where X = Cl or I). The elemental analysis confirms this composition. These are presumably four co-ordinated tetrahedral complexes.



The infrared spectral data of Cd(II) chloride and Cd(II) iodide complexes of β-4-methoxyanilinocrotono-phenylhydrazide; Hg(II) chloride complex of β-3-methoxyanilinocrotono-phenylhydrazide and Zn(II) chloride complex of β-2-methoxyanilinocrotono-phenylhydrazide and their assignments are given in Table-II.

All the spectra show medium absorption bands at ~3040 cm^{-1} and 2940 cm^{-1} due to aromatic and aliphatic -C-H stretching respectively The strong bands at 1600 cm^{-1} and 1580 cm^{-1} in the spectra of ligands can

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TABLE I

APH—metal halide complex and Molecular Formula	M.P. °C	Molecular Weight Rast's Method		Metal (%)		Halogen (%)	
		Obs.	Calc.	Found	Req.	Found	Req.
1	2	3	4	5	6	7	8
2-methoxyanilino-$C_{17}H_{19}N_3O_2CdCl_2$	>330	470	480.4	23.21	23.31	14.72	14.78
3-methoxyanilino-$C_{17}H_{19}N_3O_2CdCl_2$	>330	465	480.4	23.19	23.31	14.70	14.78
4-methoxyanilino-$C_{17}H_{19}N_3O_2CdCl_2$	>330	470	480.4	23.25	23.32	14.73	14.78
2-methoxyanilino-$C_{17}H_{19}N_3O_2CdI_2$	140	650	663.4	16.83	16.94	38.15	38.28
3-methoxyanilino-$C_{17}H_{19}N_3O_2CdI_2$	149	655	663.4	16.88	16.94	38.19	38.28
4-methoxyanilino-$C_{17}H_{19}N_3O_2CdI_2$	151	650	663.4	16.85	16.94	38.16	38.28

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1	2	3	4	5	6	7	8
2-methoxyanilino-$C_{17}H_9N_3O_2HgCl_2$	282-84	555	568.6	35,15	35.28	12.39	12.48
3-methoxyanilino-$C_7H_{19}N_3O_2HgCl_2$	240-42	550	568.6	35.19	35.28	12.41	12.48
4-methoxyanilino-$C_{17}H_{19}N_3O_2HgCl_2$	252-54	550	568.6	35.22	35.28	12.44	12.48
2-methoxyanilino-$C_{17}H_{19}N_3O_2ZnCl_2$	126	420	433.38	14.98	15.08	16.30	16.39
3-methoxyanilino-$C_{17}H_9N_3O_2ZnCl_2$	193	420	433,38	14.96	15.08	16.29	16.39
4-methoxyanilino $C_{17}H_9N_3O_2ZnCl_2$	232	420	433,38	14.99	15.08	16.31	16.39

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be attributed to the unconjugated C=O and C=N groups respectively. They shift and appear at ~1620 cm^{-1} and ~1610 cm^{-1} in the spectra of chelates. The increase in the frequency of the C=O and C=N group is probably due to the co-ordination through oxygen and nitrogen of C=O and C=N group.

There are two strong bands at ~3360 cm^{-1} and ~3240 cm^{-1} in the spectra of chelate compounds corresponding to unconjugated –N–H stretching bands, indicating that nitrogen of the hydrazide is not taking part in the chelate formation.

Moderately intense bands at ~1250 cm^{-1} in the spectra of chelates are probably due to C–H bending in plane.

In all chelates, three is also a band at ~1180 cm^{-1} corresponding to –C–N stretching.

TABLE II

Some Fundamental Vibrations (cm^{-1}) of β-arylaminocrotonophenyl hydrazides in 1:1 complexes of Cd^{+2}, Hg^{+2} and Zn^{+2}.

Compound	*ν(N–H)*	*Aromatic ν(C–H)*	*Aliphatic ν(C–H)*	*ν(C=0)*	*ν(C=N)*	*ν(C–N)*	*(C–H) in plane*
APH. $CdCl_2$ & CdI_2	3240 3300	3040	2940	1610	1600	1160	1250
APH. $HgCl_2$	3240 3300	3050	2940	1610	1600	1180	1230
APH. $ZnCl_2$	3240 3300	3040	2940	1620	1610	1130	1250

Acknowledgement

Author is very thankful to College Authorities for providing laboratory facilities and to Dr. C. M. Desai for guidance.

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POTENTIAL LOCAL ANESTHETICS PART XV

Synthesis of α-Secondaryamino—N-(ω.ω.-diarylalkyl) acetamides

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M. G. Science Institute, Ahmedabad-9.

Summary

2, 2-Diarylethyl amines and 3, 3-diarylpropyl amines were condensed with chloroacetyl chloride and the resulting α-chloroacetamides on treatment with secondary amines gave the title compounds.

Some diphenylethylamine and diphenylpropoylamine derivatives have been shown to exhibit analgesic as well as local anesthetic activity[1]. Hence it was of considerable interest to prepare acetamides containing a basic group from these amines. With this in view 2, 2-diarylethylamines and 3, 3-diarylpropylamines have been condensed with chloroacetyl—chloride and the α-chloroacetamides obtained were heated with excess of diethylamine, morpholine or piperidine to yield the title compounds.

Experimental

2, 2-Diphenylethyl amines were prepared from the corresponding 3, 3-diphenyl propanoic acid amides by Hoffmann degradation following the method of Deshpande and Nargund.[2] 3, 3-Diphenyl propylamines were prepared from the corresponding 3, 3-diphenyl propanoic acid amides by reduction with lithium aluminium hydride following the method of Black[3] et al.

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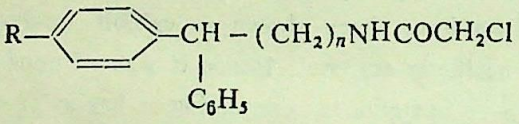
α-secondaryamino-N-(ω,ω-diarylalkyl) acetamides

The above amines were reacted with chloroacetyl chloride and the resulting α—chloroacetamides on heating with an excess of diethyl amine, morpholine or piperidine gave the title compounds as described earlier[4] in quantitative yields. The compounds prepared are shown in Table I & II.

TABLE I

α-chloro-N-(ω,ω-diarylalkyl) acetamides



No.	*R*	*n*	M.P.°C	molecular formula
1.	H	1	88—89	$C_{16}H_{16}ClNO$
2.	p-OCH_3	1	91—92	$C_{17}H_{18}ClNO_2$
3.	2, 5 (-OCH_3)$_2$	1	109—110	$C_{18}H_{20}ClNO_3$
4.	H	2	59—60	$C_{17}H_{18}ClNO$
5.	p-OCH_3	2	61—62	$C_{18}H_{20}ClNO_2$
6.	2, 5 (-OCH_3)$_2$	2	79—80	$C_{19}H_{22}ClNO_3$

Yields 75—80%

All compound analysed within allowable limits for nitrogen by Kjeldahl's (Kirk modification) method.

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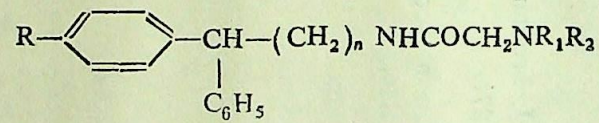
TABLE II

Secondary amino-N-(ω, ω-diarylalkyl) acetamides



No.	*R*	*n*	*NR_1R_2*	*M.P.°C*	*molecular formula*
1.	H	1	C_4H_8NO	79—80*	$C_{20}H_{24}N_2O_2.HCl$
2.	H	1	$N(C_2H_5)_2$	61—62*	$C_{20}H_{26}N_2O\cdot HCl$
3.	H	1	$C_5H_{10}N$	75—76*	$C_{21}H_{26}N_2O_0$ HCl
4.	p-OCH_3	1	$N(C_2H_5)_2$	89—90**	$C_{21}H_{28}N_2O_2$, $C_2H_2O_4$
5.	,,	1	C_4H_8NO	79—80**	$C_{21}H_{26}N_2O_3$, $C_2H_2O_4$
6.	,,	1	$C_5H_{10}N$	84—85***	$C_{22}H_{28}N_2O_2$
7.	2,5($-OCH_3$)$_2$	1	$N(C_2H_5)_2$	121—122**	$C_{22}H_{30}N_2O_3$, $C_2H_2O_6$
8.	,,	1	C_4H_8NO	100—101***	$C_{22}H_{28}N_2O_4$
9.	,,	1	$C_5H_{10}N$	89—90***	$C_{23}H_{30}N_2O_3$
10.	H	2	$N(C_2H_5)_2$	71—72**	$C_{22}H_{28}N_2O$, $C_2H_2O_4$
11.	,,	2	C_4H_8NO	83—84**	$C_{21}H_{26}N_2O_2$, $C_2H_2O_4$
12.	,,	2	$C_5H_{10}N$	81—82**	$C_{22}H_{28}N_2O$, $C_2H_2O_4$
13.	p-OCH_3	2	$N(C_2H_5)_2$	79—80**	$C_{22}H_{30}N_2O_2$, $C_2H_2O_4$
14.	,,	2	C_4H_8NO	95—96**	$C_{22}H_{28}N_2O_3$, $C_2H_2O_4$
15.	,,	2	$C_5H_{10}N$	89—90**	$C_{23}H_{30}N_2O_2$, $C_2H_2O_4$
16.	2,5($-OCH_3$)$_2$	2	$N(C_2H_5)_2$	94—95***	$C_{23}H_{32}N_2O_3$
17.	,,	2	C_4H_8NO	111—112***	$C_{23}H_{30}N_2O_4$
18.	,,	2	$C_5H_{10}N$	124—125***	$C_{24}H_{32}N_2O_3$

*hydrochloride, **oxalate, ***base Oxalates, were prepared by reacting a solution of the base in ether with a solution. of anhydrous oxalic acid in ether. All compounds analysed within allowable limits for nitrogen by Kjedahl's (Kirk modification) method.

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Acknowledgement

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'u-HOMEOMORPHISMS'

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This paper contains a redefinition of "topological map" to mean a correspondence, which includes a one to one intersection and union preserving map on the topologies as suggested by Kelley [5], page 130. Properties of such maps are studied in some detail.

The authors defined [2] a generalized unit (g. unit) in a topological space as a maximal non null set U with the property that for each open set O either $U \subset O$ or $U \cap O = \phi$. It can be seen that each topological space X is partitioned into g. units and that if x is a point in a g. unit U, then $\overline{\{x\}} = \overline{U}$.

Definition 1 : A correspondence $\underline{f} : X \to Y$ from a topological space X to a topological space Y, is said to be a u-function if for each g. unit U in X, there is a g. unit V in Y such that $\underline{f}(U) \subset V$.

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Note : By $\underline{f}(U)$, we mean the image under $\underline{f}$ of U. Such correspondence will be denoted by drawing a horizontal line below a letter.

Definition 2 : A *u*-function $\underline{f}: X \to Y$ is said to be one-one if for each pair of distinct g. units U_1 and U_2 in X, there are distinct g. units V_1 and V_2 in Y such that $\underline{f}(U_1) \subset V_1$ and $\underline{f}(U_2) \subset V_2$.

Definition 3 : A u-function $\underline{f}: X \to Y$ is said to be onto if $\underline{f}(X) = Y$.

Clearly a u-function may not be a function and a function is not a u-function in general.

If $\underline{f}: X \to Y$ is a u-function and $B \subset Y$, then $\underline{f}^{-1}(B)$ will denote the set $\{ x \in X / \underline{f}(x) \in B \}$.

Let us define "continuity", "open ness" and "closedness" of a u-function in the same manner in which these concepts are defined for function-namely we define $\underline{f}: X \to Y$ to be continuous if for each open set O in Y, $\underline{f}^{-1}(O)$ is open in X.

Theorem 1 :

(*a*) Every continuous function is a u-function, and therefore a continuous u-function.

(*b*) A one-one, onto u-function $\underline{f}: X \to Y$ is open if and only if it is closed.

(*c*) I $\underline{f}: X \to Y$ is a one-one, continuous *u*-function, then

(*i*) $\underline{g} \circ \underline{f}: X \to Z$ is a one-one, continuous u-function, if $\underline{g}: Y \to Z$ is a one-one, continuous u-function.

(*ii*) if subset F of Y is closed, then $\underline{f}^{-1}(F)$ is closed in X.

(*iii*) $\underline{f}(\bar{A}) \subset \overline{\underline{f}(A)}$ for each $A \subset X$.

(*iv*) $\overline{\underline{f}^{-1}(B)} \in (\underline{f}^{-1}\bar{B})$ for each $B \in Y$.

For a one-one u-function, conditions (*ii*), (*iii*) and (*iv*) are sufficient for its continuity.

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(d) If $\underline{f} : X \to Y$ is a u-function, then

(i) $\underline{f}$ is open if and only if $\underline{f}$ (Int. A) $\subset$ Int. $[\underline{f}(A)]$ for every $A \subset X$; and

(ii) $\underline{f}$ is closed if and only if $\overline{\underline{f}(A)} \subset \underline{f}(\bar{A})$ for every $A \subset X$.

Proof : We omit the proof of the theorem except to establish the equivalence of c (iii) and continuity of $\underline{f}$.

Since $\underline{f}$ is one-one $[\underline{f}^{-1}\ \underline{f}(A\ ]^* = A^*$ (for every subset A of X, by A^* we mean the set $\cup \{ U/U$ is a g. unit in X and $U \cap A \neq \phi \}$).

Then, because $\overline{A^*} = \bar{A}$ for every $A \subset X$;

$$\overline{\underline{f}^{-1}[\ \underline{f}(A)\ ]} = \bar{A} \qquad \ldots (1)$$

Now, Since $\underline{f}$ is continuous, $\overline{\underline{f}^{-1}\ [\underline{f}(A)\ ]} \subset \underline{f}^{-1}\ [\overline{\underline{f}(A)}\ ]$.

Then (1) implies that $\bar{A} \subset \underline{f}^{-1}\ [\underline{f}(A)\ ]$, and

hence $\underline{f}\ (\bar{A}) \subset \overline{\underline{f}(A)}$.

To prove converse, let F be a closed subset of Y.

Then $F^* = F$ and therefore $\underline{f}\ [\underline{f}^{-1}(F)\ ] \subset F$. Also

since $\underline{f}\ [\underline{f}^{-1}\ \overline{(F)}\ ] \subset \overline{\underline{f}[\underline{f}^{-1}(F)\ ]}$ is given, $\underline{f}\ [\overline{\underline{f}^{-1}(F)}\ ] \subset F$; Which ultimately implies that $\overline{\underline{f}^{-1}(F}, \subset \underline{f}^{-1}(F)$, that is $\underline{f}$ is continuous.

Definition 4 : A one-one, onto, open (closed), continuous u-function $\underline{f} : X \to Y$ is said to be u-homeomorphism. And if u-homeomorphism exists between topological spaces X and Y, then they are said to be u-homeomorphic (written as $X \cong Y$).

Theorem 2 : The following three statements are equivalent :

(a) $\underline{h} : X \to Y$ is a u-homeomorphism

(b) $\underline{h}(\bar{A}) = \overline{h(A)}$ for each $A \subset X$.

(c) $\underline{h}^{-1}(\bar{B}) = \overline{h^{-1}(B)}$ for each $B \subset Y$.

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Proof : Follows from Theorem 1.

Remark : It is easy to see that

(*a*) the relation "to be u-homemorphic" is an equivalence relation on the class of topological spaces,

(*b*) every homeomorphism is a u-homeomorphism,

(*c*) if $\underline{h} : X \to Y$ is a u-homeomorphism, then it is a one-one correspondence between

(*i*) the g.units of X and g.units of Y; and

(*ii*) the open sets of X and the open sets of Y.

(*d*) u-homeomorphism preserves the unions and intersections of open sets; and consequently it preserves the properties which are completely described by open sets (closed sets) and g. units.

As a result of Remark (*d*), we find that axioms of compactness and countability [6] (namely compactness, Lindelofness, local compactness, separability, first countability, second countability, meta compactness, para compactness, full normality, developability etc.); the axioms of connectedness [6] (namely connectedness, path connectedness, zero dimensionality, local connectedness etc.) are preserved under u-homeomorphisms. Also it can be proved that the separation axioms T_0, T_D [1] and [8], Strong T_0 [8], T_1, us [9], KC [9], T_2, T_3 and T_4 are not preserved, while regularity, complete regularity, normality, semiregularity etc. [6] are preserved. The above results are trivial as can be seen from the proof of one of these results given here.

Result : Complete regularity is preserved under *u*-homemorphism.

Proof : Let $\underline{h} : X \to Y$ be a u-homeomorphism, and let X be completely regular. Ler F be closed in Y and $y \in Y-F$. Then since F is closed,

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there is a g. unit V in Y such that $y \in V$ and $V \cap F = \phi$. And therefore

$$h^{-1}(y) \cap \underline{h}^{-1}(F) = \phi.$$

Let $f : X \to [0, 1]$ be the continuous function such that

$f(x) = 0$ for $x \in \underline{h}^{-1}(y)$

and $f(x) = 1$ for $x \in h^{-1}(F)$.

Clearly for $\underline{h}^{-1} : Y \to [0, 1]$ is a continuous function, which satisfies the requirement to show that Y is completely regular.

Final Remark :

As an extension of Kelley's remark, mentioned earlier, A. S. Davis [4] suggested a scheme of separation axioms, which he named as regularity axioms and denoted them as R_0, R_1 and R_2. Later M. G. Murdeshwar and S. A. Naimpally [7] studied R_1. The authors [3] have defined and studied unit-wise separation postulates u-T_D, u-strong T_0, u-$T_1(\equiv R_0)$, u-us u-KC, u-$T_2(\equiv R_1)$, u-T_4 and have shown that the hypotheses of T_0, T_D, Strong T_0, T_1, us, KC, T_2 and T_4 can be weakend into corresponding unit wise separation postulates. All unit-wise separation postulates are preserved under u-homeomorphisms. Finally we conclude by mentioning that a metrizable space can be found to be u-homeomorphic to a given pseudo metrizable space.

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